CONTRIBUTIONS TO THE JAPANESE ASCIDIAN FAUNA XXIX. - NOTES ON SOME CLAVELINIDS FROM THE JAPANESE WATERS-

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CONTRIBUTIONS TO THE JAPANESE ASCIDIAN FAUNA XXIX. NOTES ON SOME CLAVELINIDS FROM THE JAPANESE WATERS[1)]

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With Text-figures 1–7 and Plate I

So far seven species of the subfamily Clavelininae have been reported from the Japanese waters. Generally speaking, the zooids are structured rather similarly throughout the subfamily. Putting aside the state of colony, as a result of budding, the structure of the thorax and the stomach may offer the most useful clues to the specific identification of preserved specimens, although it seems not so difficult to make distinction between living specimens of different species, as the colour pattern is generally specific. In preserved specimens, however, the colour fades out very easily and the thorax is usually found more or less contracted unless the specimens are anaesthetized before fixation. In these specimens, therefore, it is rather difficult to read the exact number of stigmatal rows, to see the exact situation of the anus, and to trace the thoracic musculature correctly. The last may look quite differently according to the degree of contraction. Moreover, the feature of the stomach seems to differ considerably according to the physiological state of the zooids in respective colonies, e.g. the grade of maturity of gonads. All these can be the causes of bringing about some taxonomical confusions at the generic or specific level.

Recently the last author had a chance to examine a nicely preserved large colony of a clavelinid which was collected by Mr. Shohei Shirai in the southern part of Mié Prefecture on the east coast of Kii Peninsula and the first author examined several colonies of clavelinids which were collected by Dr. Shin'ichiro Fuse off the southeastern coast of Tokusima Prefecture, Sikoku Island and by Mr. Torao Yamamoto off Minabe about 5.6 km north of the laboratory. The former was identified with *Clavelina coerulea* Oka, while the colony from Minabe was attributed to *Dendroclavella elegans* Oka and those from Tokusima were found to be a new species. In these studies, the types of *C. coerulea* Oka and *Dendroclavella elegans* Oka were checked carefully and the colonies of "*Clavelina fasciculata* by Tokioka" from Sagami Bay were reexamined. Throughout these examinations considerations were paid to the different features of some structures presented by zooids of the same species in differ-

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ent states of preservations, as some comments are given on them at the end of this paper.

Before going further, the authors wish to express hearty thanks to Mr. S. Shirai, Dr. S. Fuse and Mr. T. Yamamoto for their kindness in offering these interesting specimens for the present studies, and especially to Prof. Hiroshi Watanabe, the director of the Shimoda Marine Biological Laboratory of Tokyo Kyoiku University, for the loan of precious types of *Clavelina coerulea* and *Dendroclavella elegans*. The authors also acknowledge with their cordial thanks the priviledge of reexamining the colonies of "*Clavelina fasciculata* by Tokioka" from Sagami Bay, which are kept at the Biological Laboratory, Imperial Household.

Clavelina coerulea Oka, 1934

(Pl. I above, and Figs. 1 and 2)

Oka, A. (1934): Proc. Imp. Acad., vol. 10, no. 6, pp. 365–366.

A large colony including about one hundred zooids crowded on a common base which is 40 mm × 30 mm in extent and consisting of creeping stolons branched and entangled. It was collected by Mr. Shohei Shirai from an exposed sublittoral rocky reef around Tusima Island on the southeastern coast of Kii Peninsula and offered to the last author together with a transparency in natural colour showing its living state.

Zooids are up to 25 mm in length, all issued from the creeping stolons but never presenting any fusion between zooids (fig. 1, A). The very thin surface layer of the gelatinous test is rather hard but the inner part, in which the zooid is embedded, is very soft; while the stolons are sufficiently hardened on the surface just like the hydrozoan periderm. In the preserved state, the thoracic portion is nearly as long as the abdominal portion which is then supported by the peduncular portion about half as long as the abdominal portion. The atrial siphon terminal and the branchial subterminal, both siphons very short and each with a round aperture. The mantle body of fully grown zooids is usually 13 to 14 mm in length, excluding the posterior process connecting the posterior end of the abdomen to the stolon. The thorax is as long as the abdomen. When alive (Pl. I), the whole zooid is coloured cobaltblue, especially deeply in the anterior part and over the incubatory pouch of the thorax, in addition there is a thin darker line along the left side of the distal portion of the rectum. In preserved specimens, the colour of the deeply pigmented portions mentioned above is turned to purplish, while the abdomen is quite whitish.

Thorax: There are 20 to 30 muscles, nearly transverse but slightly slanting to the dorsal side. In addition to these, there are a few to several longitudinal muscles converging to the atrial siphon and running outside the transverse ones, of which the ventralmost one is much longer than the others, reaching posteriorly to the middle of the thorax (fig. 1, B) or further to the posterior end of the thorax (fig. 2, B). There are 16 to 18 stigmatal rows, usually one more row on the right side; up to 45 stigmata in each row. Transverse vessels are provided each with a well developed membrane.

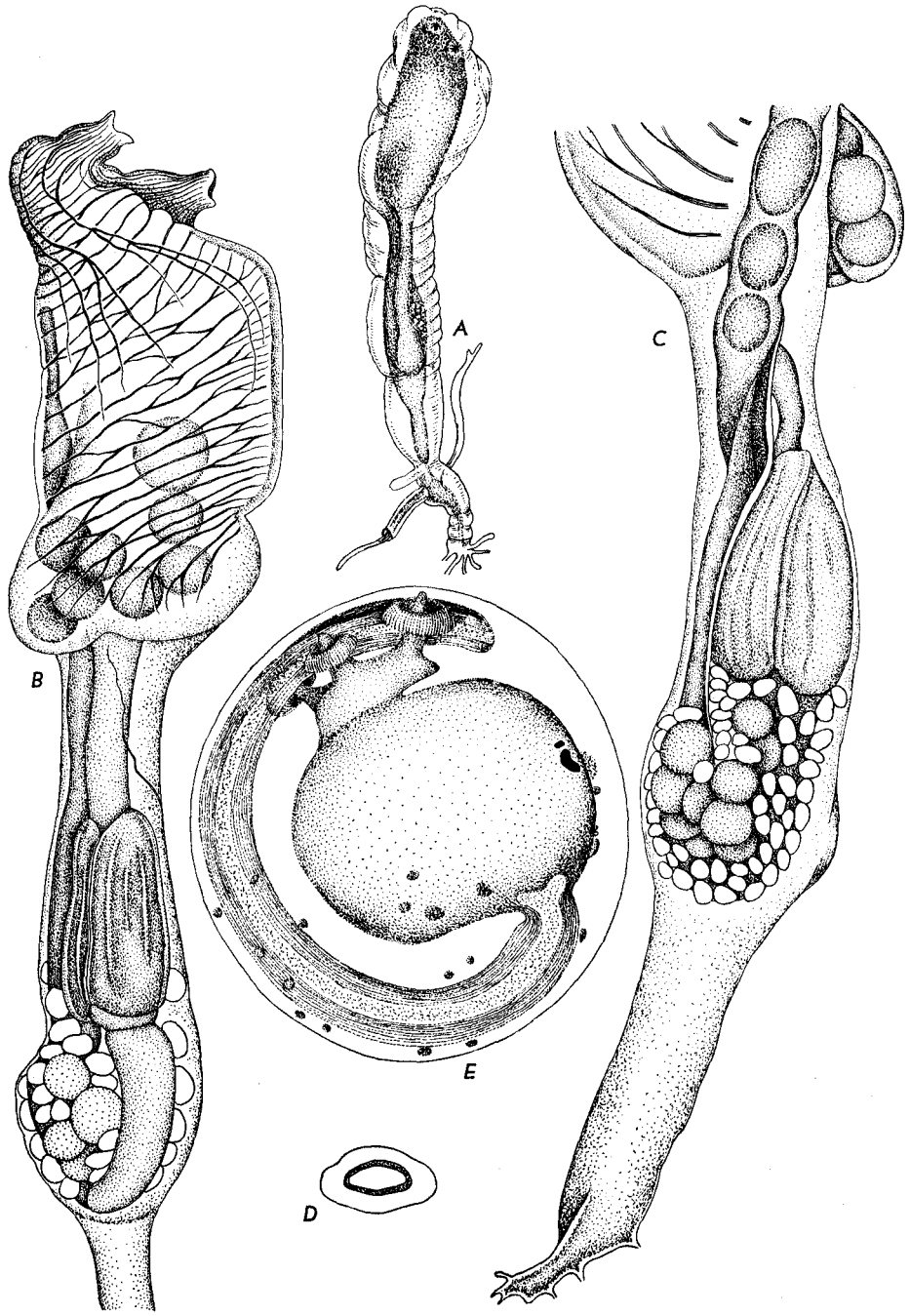

Fig. 1. *Clavelina coerulea* Oka, from Mié Prefecture. A: Zooid. B: Mantle body, right side. C: Abdomen, left side. D: Ciliated groove. E: Embryo in advanced stage.

Embryos and fertilized eggs up to 12 in a total in the incubatory pouch of a 6 mm long thorax or 18 in a total in a 7 mm long thorax. The rectum in the thorax contains about 5 faecal pellets and is attached to the branchial sac around the level of the 13th transverse vessel, but leaving the distal portion of a considerable length free in the atrium; therefore the anus originally opening in the posterior part of the thorax in the live state in which the stigmatal rows are all fully extended, may be situated much anteriorly, for instance at the anterior third, in more or less contracted preserved specimens in which the anterior stigmatal rows, not supported by the rectum wall, are all much decreased in height. Anal margin plain. The tentacular ring consists generally of 6 larger and 6 smaller tentacles, one or two minute ones may occur in their intervals. The ciliated groove (fig. 1, D) is a small elliptical opening, slightly elongated transversely. Dorsal languets are nearly median.

Abdomen: A pair of abdominal longitudinal muscle bands consist respectively of about a dozen fibres; one is running along the rectum, while the other is extended on the ventral side to cover the stomach. The stomach occupies roughly the middle third of the abdomen and is furnished with several striations in addition to the typhlosole. These striations, some of them are shorter, are not definable on the external surface, though distinctly represented by wall thickenings on the internal surface of the stomach. The hind-stomach is short and passes to the long middle-intestine through the boundary not remarkable externally but definable by different contents in respective portions. The middle-intestine passes to the rectum through a faint constriction at the posterior end of the abdomen. The gonads occupy the left side of the intestinal loop posterior to the pyloric end of the stomach, the ovary is central and testicular follicles are distributed peripherally.

Ovarian eggs are 540 μ in diameter at the maximum, while fertilized eggs in the incubatory pouch in the thorax are 600 to 720 μ, and embryos are 960 to 1140 μ in diameter in examined specimens. In embryos, the distal end of the tail is folded and held between two and one of the attachment processes; the stalk of processes is fully extended in the 1140 μ long embryo (fig. 1, E), but still short in the 960 μ long one. Two pigment flecks are arranged antero-posteriorly near the middle of the trunk; small white speckles are distributed sparsely in the posterior half of the embryonal capsule.

Remarks: The type (M725: 278 in Zool. Inst., Tokyo Kyoiku Univ.) of the species was collected by the late Dr. Taku Komai at Sibusi in Kagosima Prefecture on April 10, 1934. The zooids were coloured beautifully cobaltblue, especially deeply around the apertures and the anus opening on the left side of the dorso-median line in the posterior part of the atrium. The fine muscle fibres are running obliquely on the thorax. There are 16 stigmatal rows. The stomach surface is smooth. The features mentioned above are quoted here from Oka's original description of the species to show some problematical points in the identification of the present specimen. At a glance a close resemblance in colouration in the living state is evident between the Oka's type and the present specimen (Pl. I, above). The number of stigmatal rows conforms rather well to each other. The situation of the anus is much anterior in

the present specimen, but that in the living state confirmed on the photograph in natural colour by a deep blue spot is evidently found in the posterior portion of the thorax as given in the original description. However, the exact comparison of the thoracic musculature and the stomach wall had been impossible on the original description. Therefore, the type colony was reexamined on these features.

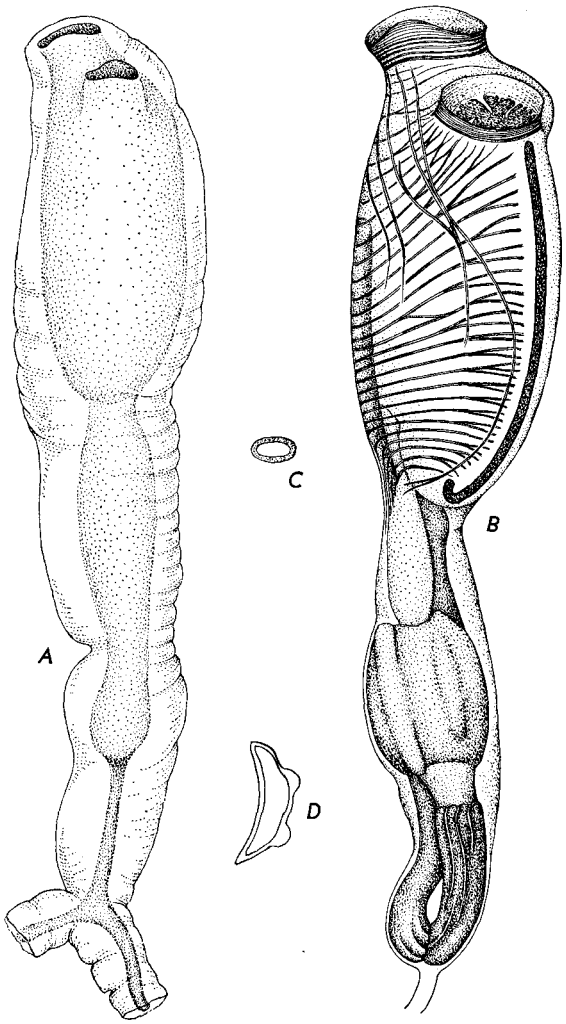

Fig. 2. *Clavelina coerulea* Oka, type specimen. A: Zooid. B: Mantle body, right-ventral side. C: Ciliated groove. D: Section of stomach.

The type colony consists of about 35 zooids, which are 10 to 20 mm long, and contains in stolons a number of small roundish ampullae, or rudimentary buds, around the base of some zooids. A 14 mm long zooid and a 18 mm long one were

dissected to confirm the internal structures; their mantle bodies are 10.5 mm and 14 mm long respectively. In both zooids, the gonads are quite immature. It was found that the thoracic musculature of the larger zooid (fig. 2, B) was quite the same as that in the present specimen from Mié Prefecture and the stomach surface was marked with 3 prominent folds (fig. 2, D) in both zooids. Some other features in these zooids are recorded below for future studies.

After the preservation in formalin for about 40 years, a purplish blue tint remains still in the dorsal half of the atrial aperture. The thorax is nearly as long as the abdomen. About a dozen fine sphinctors around the branchial and atrial apertures. The anus is situated approximately at the level of the anterior third of the branchial sac and its margin is wholly plain; the distal portion of the rectum is left free in the atrium in a considerable length. There are 15 to 17 stigmatal rows and two of them are stretched posterior to the oesophageal opening. Fifty or more stigmata in each row. Transverse vessels are provided each with a well developed membrane. Dorsal languets are nearly median. Tentacles consist of 6 larger and 6 smaller ones, alternating regularly. The ciliated groove (fig. 2, C) is a small elliptical opening slightly elongated transversely. The stomach occupies the middle third of the abdomen. The hind-stomach and the middle-intestine are defined very clearly (fig. 2, B); the latter is marked with a few striations and jointed to the rectum at the posterior end of the abdomen. The beginning end of the rectum is furnished faintly with several short plications.

Throughout the features observed on two zooids of the type colony, it is quite evident that the present specimen is identical with *C. coerulea* Oka. Especially, the coincidence in the peculiar feature of the thoracic musculature seems to support this identification decisively. Also it is cleared that the stomach is essentially folded in *C. coerulea* and therefore the species belongs clearly to the genus *Clavelina*.

Clavelina elegans (Oka, 1927)

(Figs. 3–5)

Dendroclavella elegans Oka, A. (1927): Proc. Imp. Acad., vol. 3, no. 8, 555–557, figs. A and B.
Clavelina fasciculata (non Van Name, 1945) Tokioka (1953): Ascidians of Sagami Bay, 202–203, figs. 1–3 in pl. 27.

A single colony was found by Mr. Torao Yamamoto in the organisms caught by gill nets for spiny lobster fishery set off Sakai in Minabe, Wakayama Prefecture, about 20 m deep, on March 8, 1975. The colony consists of four club-shaped zooids born on a common peduncle just as in *Dendroclavella elegans* Oka or the typical *Clavelina fasciculata* described by Van Name. The test of the zooid is thin, transparent, but somewhat tough; while on the peduncle it is thick, translucent and somewhat hard in the distal half, but very hard, opaque and coloured pale orange in the proximal half. Zooids are 20 to 22 mm long and about 9 mm wide; the peduncle is about 14 mm in length.

Zooids: The thorax is longer than the abdomen in extended zooids, though the former may be shorter than the latter in contracted ones. Both apertures are very large and margined plainly.

Thorax: There are 5 or 6 longitudinal muscles on each side of the thorax. Twenty-six to 30 stigmatal rows are present, each containing 60 to 70 stigmata. Tentacles 24, inclusive of large to small ones. The ciliated groove is a longitudinal slit. The

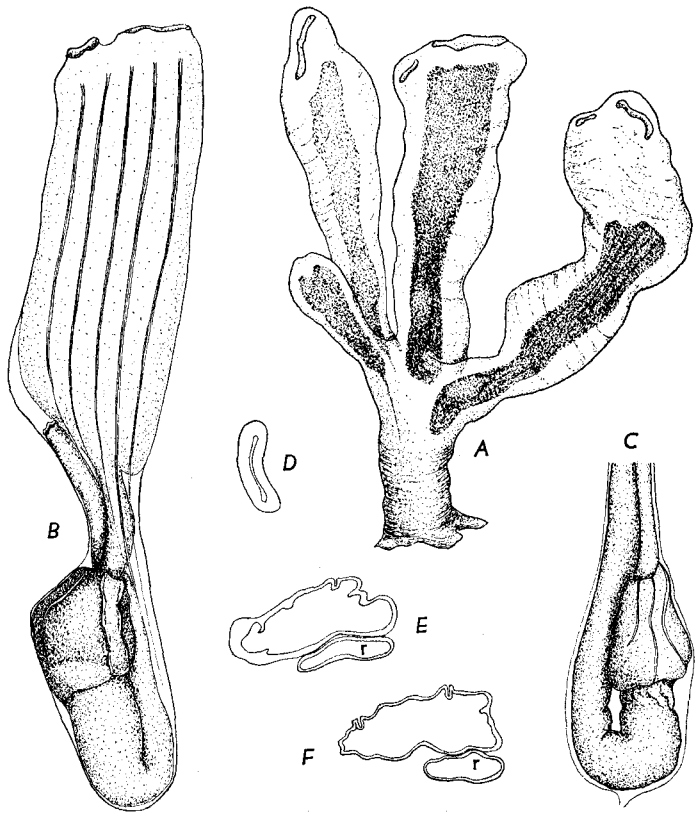

Fig. 3. *Clavelina elegans* (Oka), from off Minabe, Wakayama Prefecture. A: Colony. B: Mantle body of 22 mm long zooid, right side. C: Abdomen of 19 mm long zooid, left side. D: Ciliated groove. E: Optical section of stomach, 22 mm long zooid. F: The same, 8.5 mm long zooid. r: rectum.

anus opens at the level as high as the posterior three stigmatal rows and its margin is divided into two lobes which are then subdivided into several lobules.

Abdomen: The stomach occupies the middle one-third of the abdomen and is marked with 2 or 3 longitudinal plications on the surface; the hind-stomach is not so distinct. No gonad was found in any zooid.

Remarks: Evidently only the fasciate feature of the colony can never be a decisive specific criterion to any clavelinids. In the present identification, therefore, the ex-

istence of longitudinal thoracic muscles, the ciliated groove as a longitudinal slit, and the stomach furnished with a few longitudinal plications were checked between the present colony and some already described forms.

In the original description of *Clavelina fasciculata,* no details are given as to the thoracic musculature and the ciliated groove, though it is stated that "The few specimens of *Clavelina* from the Gulf of California which I have seen appear to belong to a species allied to *C. huntsmani,* having zooids of the same structure as in that species (including the four-ridged stomach), ···" (Van Name 1945, p. 140). As *C. huntsmani* Van Name, 1931 is furnished mainly with longitudinal muscles on the thorax (Van

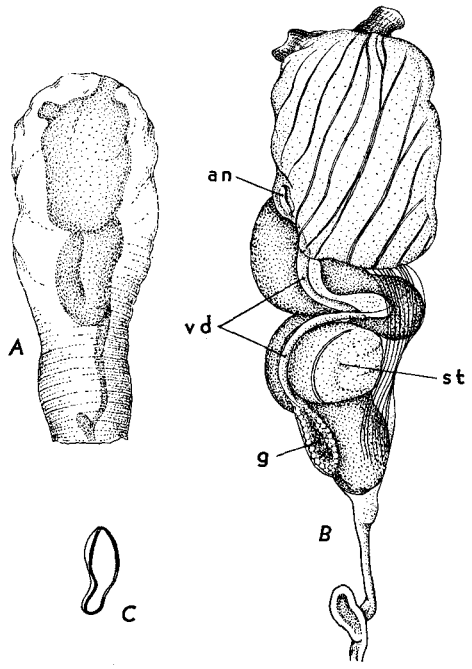

Fig. 4. *Clavelina elegans* (Oka), neotype from Sagami Bay. A: Zooid. B: Mantle body, right side. C: Ciliated groove. an: anus. g: gonad. st: stomach. vd: vas deferens.

Name 1945, p. 141) and provided with the dorsal tubercle that is longitudinally elongate oval and with an orifice of similar form (p. 142), the present colony is regarded to share with *C. fasciculata* quite the same features of these structures. However, there are more stigmatal rows in the present colony than in *C. huntsmani* in which 16 to 20 rows, with often about 40 stigmata in each, are given; this difference seems rather significant, though might be merged in an intraspecific variation.

On the other hand, the description made by Tokioka (1953) of *C. fasciculata* from Sagami Bay lacks the explanation of the thoracic musculature, and further in those colonies the zooid is provided with the ciliated groove transversely elongate.

In order to explain these points fully, the colonies from Sagami Bay were reexamined. A 12 mm long zooid (fig. 4) of a colony in Sp. No. 137 (Proto. 240) kept at the Biological Laboratory, Imperial Household, was dissected and observed in detail. The gelatinous test is very thin and transparent in the distal half around the zooid, but becomes posteriorly thicker and less transparent in proximal peduncular portion; the surface is smooth. The mantle body (fig. 4, B) of the zooid is 8.2 mm long in a strongly contracted state, and the thorax and the abdomen are nearly equal in length in this state. There are 4 to 6 longitudinal muscles converging to the branchial siphon and 3 or 4 ones ending towards the endostyle, all these muscles seem rather oblique on the contracted thorax, but may look longitudinal in a fully extended state, except for a few finer ones along respective median lines. They are extended posteriorly into the ventral muscle band on the abdomen, that consists of about a dozen muscles. There are 16 stigmatal rows and up to nearly 80 stigmata in each, though 21 rows were recorded previously (1953). The ciliated groove is a longitudinal opening (fig. 4, C). The groove was given (1953) as a crescent slit and shown laid transversely in the figure (figure 3 in pl. 27). As it is seemingly impossible that the ciliated groove assumes a longitudinal slit in some zooids but a transverse opening in some others, the configuration given previously may be a mistake of drawing the groove as it was seen when the area including the groove was observed being laid transversely under the microscope. The anus is bilobed and open a little above the dorso-posterior corner of the thorax.

The stomach is globular and its surface is quite smooth, but with a very distinct typhlosole. On the inner surface of the stomach wall, however, one or two longitudinal ridges are definable faintly on each side of the typhlosole; these may be more prominent in the pyloric half than in the cardiac portion. Therefore, in a strict sense the stomach can never be mentioned as smoothly surfaced. The hind-stomach is defined but indistinctly, and very short. Zooids in the colonies collected from Sagami Bay, 70 m deep, on November 13, 1950 are matured.

Excepting that more stigmatal rows are found in the colony from off Minabe, that colony resembles the colonies from Sagami Bay redescribed just above rather than *Clavelina fasciculata* Van Name from the Gulf of California, especially in the feature of the thoracic musculature and the ratio in length of the thorax to the abdomen. The ratio should be much smaller in *C. fasciculata* and *C. huntsmani,* because the structure of the former resembles closely that of the latter, in which the abdomen is much longer than the thorax (cit. Van Name 1945, fig. 67A on p. 142). Comments are to be given later as to the structure of the stomach wall in Japanese species.

The reexamination of the colonies from Sagami Bay reminded the authors of *Dendroclavella elegans* that had been described by Oka in 1927 as a new species of a new genus on the colonies collected in Sagami Bay off Matuwa and Tateyama. This new genus was characterized by a fasciate feature of the colony, while the specific characteristics were not given sufficiently to define this form distinctly. No words are paid as to the thoracic musculature and the ciliated groove. However, 18 stigmatal rows are recorded, the stomach wall is smooth, and only the last part of the rectum is pro-

truded into the thorax. All these features are shared with the colonies of "*C. fasciculata* by Tokioka" from Sagami Bay. Then, a question may come quite naturally into consideration if "*C. fasciculata* by Tokioka" is identical with *D. elegans*.

The type colonies (M499: 313 in Zool. Inst., Tokyo Kyoiku Univ.) of *D. elegans* was reexamined, but unfortunately they were already in a heavily deteriorated state; the internal structures of the thorax had already disappeared wholly and the alimentary canal was definable only by muddy contents held in a thin membranous structure. However, up to probably 8 oblique striations were defined on each side of the emptied thorax (fig. 5), and these may be the traces of muscles decayed out.

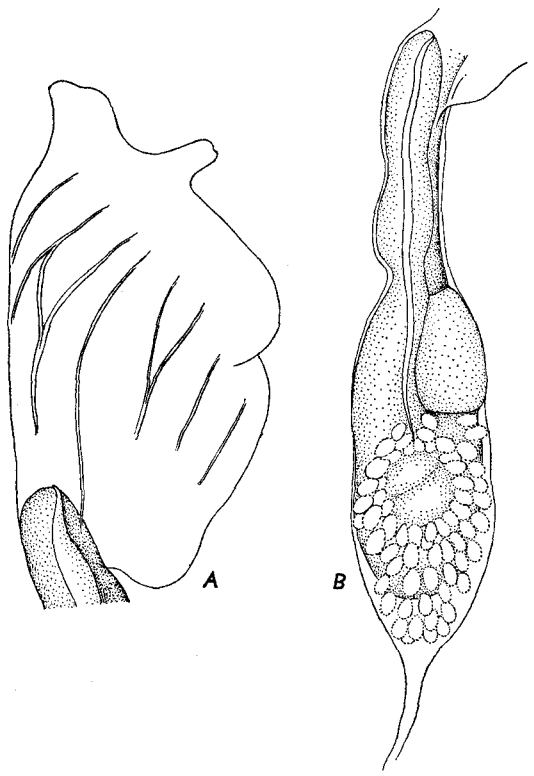

Fig. 5. *Clavelina elegans* (Oka), deteriorated type. A: Probable traces of thoracic muscles. B: Outline of alimentary canal.

The stomach, that is quite smooth at least on the membranous structure, is situated at the middle of the abdomen. These features seem to suggest the identity of the two forms in question from the same locality referred to above.

Speaking very strictly, *D. elegans* may be put aside forever as an uncertain species because of its insufficient specific description. However, it may be a way to regard the previous and present descriptions of "*C. fasciculata* by Tokioka" from Sagami Bay as the redescription of *D. elegans* and to designate the Sp. No. 137 (Proto. 240) of the Biological Laboratory, Imperial Household, for the neotype of *D. elegans*. The

authors would like to take the last way to diminish confusions even a little in the taxonomy of Japanese clavelinids.

The main features of *Clavelina elegans* (Oka, 1927) may be summarized as follows: Colony is fasciate. Thoracic muscles are up to 10, main ones converging to the branchial siphon are longitudinal rather than oblique in an extended state. From 16 to 21 ~ 26 to 30 stigmatal rows, 60 to 80 stigmata in each. Ciliated groove is a longitudinal slit. Stomach is situated at the middle of abdomen; the external surface is smooth, but in addition to typhlosole there are a few ridges on the inner surface and the wall may be folded probably along these ridges when strongly contracted.

Clavelina obesa n. sp.

(Fig. 6)

The material was collected by Dr. Shin'ichiro Fuse from the rocky slope, not so much exposed and about 10 m deep, in an inlet of Takegasima Island off the coast of Tokusima Prefecture facing the Kii Channel on August 3, 1973. According to the colour transparency made by the collector himself and his statement, some colonies of probably the same species, each consisting of 15 to 20 zooids, were found to and fro in that area. And when alive, the zooids were shining in bluish white through the transparent test and looked at a distance as if they were luminescent. The material contains 23 zooids. The colony is completely social; zooids are perfectly independent and connected one another only by stolons.

Zooids: They are rather stout, 11.5 to 21 mm in length and 5 to 7 mm in width. No peduncle is definable. The test is gelatinous, quite colourless and transparent, though less so in the proximal part. The test surface is smooth, but with minute papillae around both apertures. The thorax is a little longer than the abdomen; the former attaining 1.3 times as long as the latter in some zooids.

Thorax: The atrial aperture terminal, while the branchial subterminal and opened dorsads; each opening on a short siphon and smoothly margined. There are 10 to 14 thick oblique thoracic muscles on each side, slanting gently from the anteroventral to the posterodorsal side of the thorax. Fourteen to 15 stigmatal rows, each containing 60 to 75 stigmata. Tentacles about a dozen, partly in regular alternation of larger and smaller ones. The ciliated groove is a simple longitudinal slit. (fig. 6, D). The anus is situated posteriorly, with 3 or 4 stigmatal rows between it and the posterior end of the thorax, and divided into two plain lobes.

Abdomen: The stomach is superficially situated in the anterior half of the abdomen and nearly half as long as the proximal branch of the intestinal loop. Its surface is furnished with four longitudinal plications which consist of a typhlosole and three epithelial infoldings (fig. 6, E, F above), though in some larger specimens some of these plications may become much less distinct and almost obscure, being represented by thinning of the stomach wall (fig. 6, F below). The hind-stomach is short and may be indistinct in some zooids; the middle-intestine indistinct. Reddish brown eggs and pinkish white testicular follicles are packed in the posterior half of the abdomen

on the lateral side of the intestinal loop; ovarian eggs are about 700 μ in diameter. Some eggs are found discharged into the incubatory portion of the atrium.

Remarks: Apart from the colouration in the living state, among the clavelinids described so far from the western part of the North Pacific *Clavelina cyclus* Tokioka and Nishikawa from the waters of Okinawa resembles most closely the present specimens. Both forms share the following features: a rather stumpy body shape of zooids, with the thorax frequently larger than the abdomen even in the preserved state; the thoracic

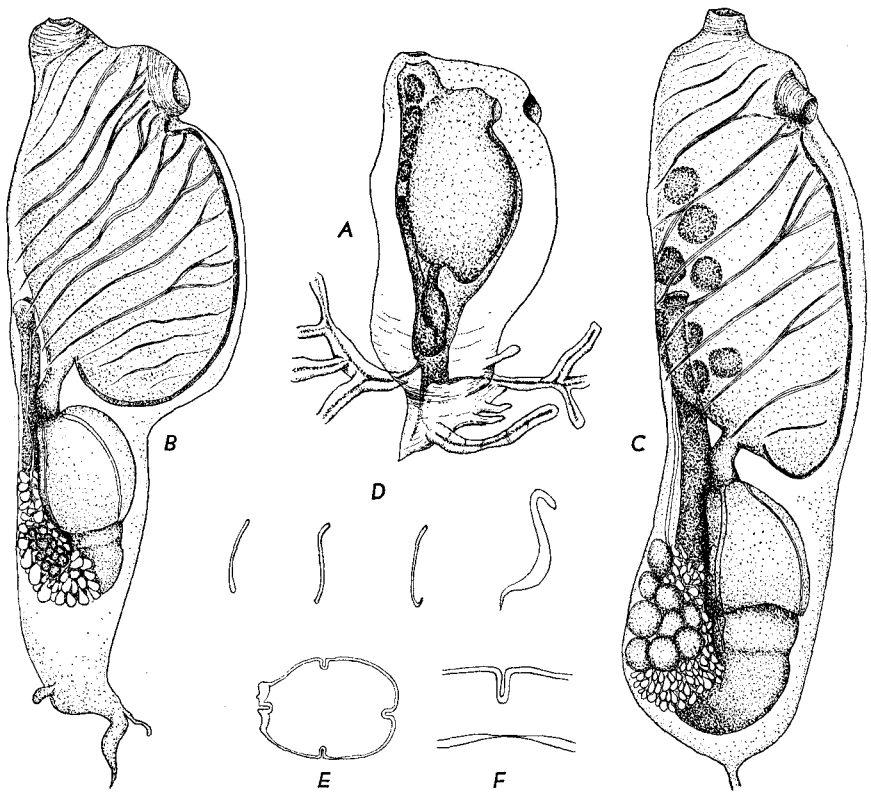

Fig. 6. *Clavelina obesa* n. sp., from off Tokusima Prefecture. A: Holotype. B: Mantle body of holotype, right side. C: Largest mantle body, right side. D: Ciliated grooves of 13.5 to 21 mm long zooids. E: Optical section of stomach. F: Infolding of stomach wall (above) and the same site represented by thinning of wall in dilated stomach (below).

musculature consisting of up to about 15 oblique muscles, the existence of 14 to 15 or 15 to 22 stigmatal rows, the ciliated groove as a longitudinal slit, the anus opening at or near the dorsoposterior corner of the thorax, and the situation and the plicated feature of the stomach. However, the thoracic muscles in the main part of the thorax in the present specimens are seemingly considerably fewer than in *C. cyclus*, generally about 10 or less, and moreover they are rather transverse, though slanting down dorsads; while in *C. cyclus* at least several dorsal muscles are converging to the branchi-

al siphon and therefore rather longitudinal than oblique. The number of stigmatal rows is continuous between the two forms, but the range is 15 to 22 in *C. cyclus* while 14 to 15 in the present specimens; further stigmata in a row are 20 or a little more in the former but much more, 60 to 75, in the latter. In addition to these differences, the remarkable difference in the colouration or colour pattern in the living state may probably be decisive.

Of exotic species, *Clavelina dellavallei* (Zirpolo) from the Mediterranean Sea seems to be related closely to the present specimens in the size and feature of zooids, the thorax larger than the abdomen, the oblique (according to fig. 4 in Salfi, 1931) ~ longitudinal (Pèrés, 1956) thoracic musculature, the existence of 14 stigmatal rows (Pèrés, 1956) and in the lower situation of the anus. However, *C. dellavallei* is almost solitary (Zirpolo 1925, fig. 1) and provided with only 5 (Pèrés, 1956) or 6 (according to fig. 4 in Salfi, 1931) thoracic muscles; these are roughly half as many as in the present specimens. Moreover, this species is coloured grayish blue as in *C. cyclus* when alive. Throughout the above given comparison, it is concluded that the present specimens represent a new species, which is here named by both authors *Clavelina obesa* for its short and somewhat fat appearance of the zooid. The minutely papillated feature of the test around the anterior end of the zooid can probably be a character peculiar to the present new species, though it is not known whether or not the test is really completely smooth in the same part of the zooid in all other known species.

Clavelina cyclus Tokioka and Nishikawa, 1975

(Pl. I below, Fig. 7)

Tokioka and Nishikawa (1975): Publ. Seto Mar. Biol. Lab., 22 (5), 326–331, figs. 7–20.

Two additional colonies of this recently established species were supplied to the authors by Prof. Kiyoshi Yamazato, Ryukyu University, who collected them on January 1, 1976 from the shallow water, 5 m deep, by the Sesoko Marine Biological Laboratory of the University, situated on Sesoko Island, Okinawa. The authors express their hearty thanks to Prof. Yamazato for his generosity in submitting the specimens to them.

The colonies consist respectively of 20 and 40 fully matured zooids crowded on a common basal mass, up to 5 mm in thickness. This state rather resembles that of the paratype, but shows clearly a more compact feature: only the distal 8 to 13 mm long portions of zooids are separated, while the proximal parts of zooids are embedded in most part of the colonies by the common test as seen in fig. 7, A, though a few wholly independent zooids are seen along the periphery of the colonies. The proximal-most portion of the type zooid, which is described as the peduncle, is better mentioned as the stolon.

Zooids as the mantle body are rather smaller, about 12 mm long in somewhat contracted state. However, as the thorax is fixed in more extended state, some structures are definable more exactly. The thoracic muscles are described as oblique in

the original description, but in the present specimens more extended than the type specimens, the muscles are longitudinal rather than oblique except finer ventro-posterior ones. It may be said safely that at least the main thoracic muscles, 10 to 14 in the zooids examined, converged to the branchial siphon are longitudinal in living or finely preserved specimens. From 40 to 50 stigmata in each of 15 to 19 rows;

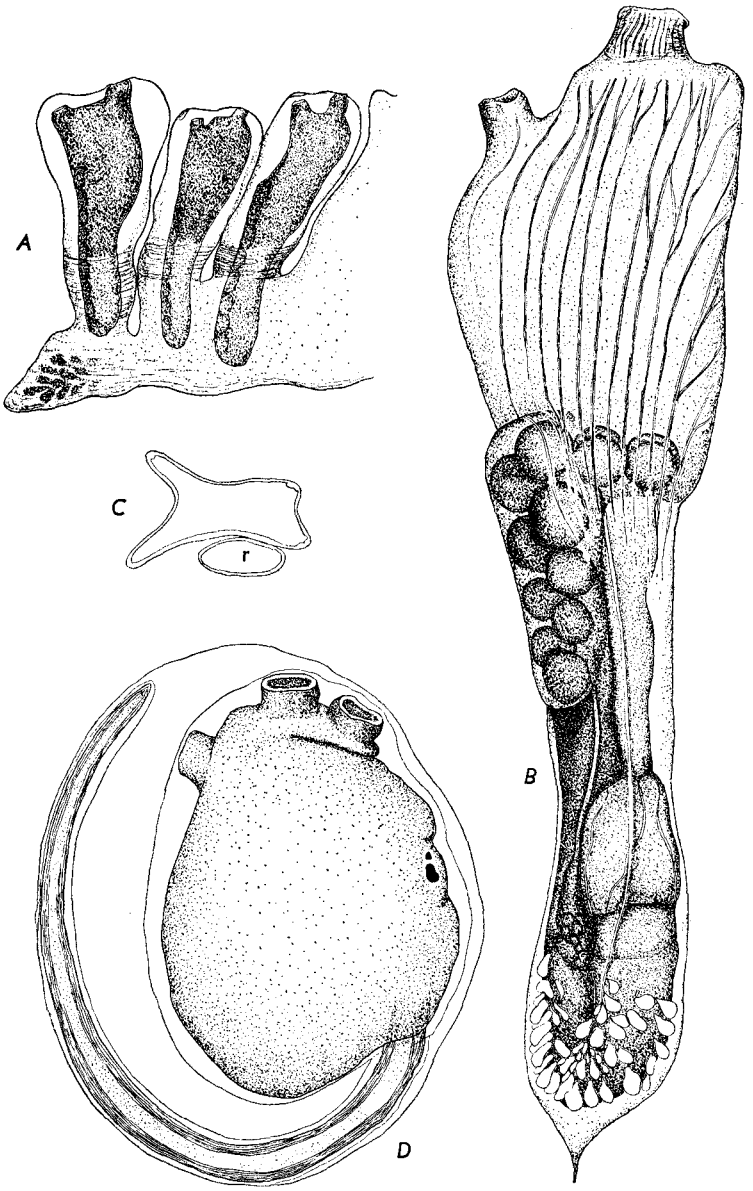

Fig. 7. *Clavelina cyclus* Tokioka and Nishikawa, from Sesoko Island, Okinawa Prefecture. A: Part of colony. B: Mantle body of 11.9 mm long zooid, right side. C: Optical section of stomach. D: 1200μ long embryo. r: rectum.

the original description giving only "20 or a little more" stigmata in a row, is made on an evident underestimate caused by strong contraction of the thorax and should be corrected to be 40 to 50. A prominent incubatory pouch is formed in an examined zooid, it is stretched from the right dorsoposterior part of the thorax posteriorly to the level of the anterior third of the abdomen and contains about a dozen of orange embryos, though it is uncertain whether the situation of the pouch is so in the live specimen. The anus opening at the dorsoposterior corner of the thorax is bilobed, or the margin may be folded into fine lobules.

The stomach is furnished with 4 distinct longitudinal ridges which are definable clearly by their translucent and colourless texture against the general wall coloured orange. The zooids retained some of their colouration in living state about a month after the preservation in formalin. The thorax was dark bluish purple, with the characteristic yellow circle around the base of the branchial siphon and generally with a yellow longitudinal band along the endostyle and/or mid-dorsal line.

Embryos are 900 to 1200 μ in diameter, with the tail end held between anterior two and posterior one of the attachment processes. In a 900 μ long embryo, the stalk of processes is not yet definable, though in a more developed 1200 μ long one the anterior protuberance supporting the attachment processes is just differentiating (fig. 7, D). One of 1200 μ long embryos is fully developed, and with elongate stalks and the well defined anterior supporting protuberance, on which, however, are found unusually 6 processes.

Considerations

Although some of the genera so far described in the subfamily Clavelininae are still questioned about their validity, generally the following 5 genera are recognized:

Genus *Archiascidia* Julin, 1904
Genus *Clavelina* Savigny, 1816
Chondrostachys Macdonald, 1858
Stereoclavella Herdman, 1890
Rhodosona Van Name, 1902
Bradiclavella Zirpolo, 1925
Dendroclavella Oka, 1927
Subgenus *Synclavella* Caullery, 1900
Synclavelina Ritter, 1903
Genus *Podoclavella* Herdman, 1890
Genus *Pycnoclavella* Garstang, 1891
Genus *Nephtheis* Gould, 1856
Oxycorynia von Drasche, 1882

Of these, only *Clavelina* and *Podoclavella* have been represented from the Japanese waters. Putting the exotic genera aside, because enough specimens are inaccessible, the most important problem in the Japanese clavelinids may be the distinction between

Clavelina and *Podoclavella*. The only difference between these genera is that the stomach wall is plicated longitudinally in the former, while quite smooth in the latter. Therefore, the stomach surface has been checked up in all Japanese described species. The plications are very distinct in *C. minuta* and *C. miniata*, both in the immature state and without any developed gonads. On the other hand, the stomach surface is quite smooth in *P. polycitorella* in the fully mature state. In the holotype of *C. cyclus*, that is provided with matured gonads but including no fertilized eggs or embryos in the thorax, the stomach surface is furnished with 4 distinct and 1 indistinct plications (Tokioka and Nishikawa 1975, fig. 15), while in the paratype with rudimentary gonads the stomach surface is marked rather vaguely with several indistinct plications (Tokioka and Nishikawa 1975, fig. 20). As a number of ampullae are included in the common basal mass of the paratype colony, it seems that the zooids have entered the retrogressive stage already and the gonads are not rudimentary but vestigial, and roughly saying two striations definable on the stomach surface may probably represent both edges of single folds. In additional specimens described in the present paper, in which some embryos are found, the stomach plications are definable only by a difference in colouration (p. 77), though the surface is seemingly smooth. In *C. obesa* the stomach looks superficially smooth, but some folds are seen clearly in the wall section (fig. 6, E, F). *C. elegans* was originally described to bear the stomach with a smooth surface, but in its neotype the stomach wall is slightly thickened longitudinally at some sites else than the typhlosole (p. 71). As the fasciate state of the colony can never be a generic criterion, the genus *Dendroclavella*, with the type species *D. elegans*, should be a synonym of *Clavelina* rather than of *Podoclavella*. In *C. coerulea*, which was also described to bear the stomach with a smooth surface, the stomach is provided with distinct plications in the immature type specimen (fig. 2, D), while marked only with several indistinct striations in fully matured specimens including some embryos inside the thorax (fig. 1, B, C); in the latter case the stomach surface may be mentioned superficially smooth.

Throughout these observations seemingly two trends are perceived. One is the order in the distinctness of the stomach plications, namely a series from *C. miniata* and *minuta* (very distinct)→*C. cyclus*→*C. coerulea*→*C. obesa*→*C. elegans* to *P. polycitorella* (wholly smooth). The plications in *C. miniata* are as clear as those in styelids, but the feature in *C. cyclus* and some others reminds ones of that of the stomach in some species of *Ascidia*. Thus, it is very difficult to judge whether the stomach described as smooth in many species is really devoid of any longitudinal ridges on the inner surface or with some slight longitudinal wall thickenings and then the wall may be folded along these when strongly contracted. Anyhow, the plications must be very few in clavelinids. There are a considerable number of species showing a stomach section with four ridges (rectangular or vierkantig). Of these ridges one should be represented by the typhlosole and two by respective edges of the stomach wall facing the rectum, therefore only a single ridge is the real plication on the free surface of the stomach. Generally a single plication is found on one or each side of the typhlosole and in some states the single plication may be represented by two slight ridges, thus several less

prominent ridges may be found on the free surface of the stomach in some specimens. The typhlosole is generally marked distinctly with two ridges, though some irregular additional ones may be seen near the pyloric end of the stomach.

The other trend concerns the changes of the stomach feature according to the physiological or maturation state of the zooid. Generally in clavelinids, budding becomes prominent with the resorption of zooids after the closure of sexual reproduction. Therefore, it is probable that the alimentary canal may start to degenerate at some level of gonadal maturation. As the stomach plications should be related with the glandular tissue, the plications might become less distinct with the decrease of the gut activity. If these trends are admitted generally, it will then be very difficult to separate strictly *Podoclavella* and *Clavelina* from each other. Even if the genus *Podoclavella* is defined as being wholly devoid of any kind of longitudinal structures on the stomach wall throughout the whole life stages of the zooid, it will be practically impossible to confirm this by only the external examination; it will need a close examination of stomach sections. However, the present authors are hesitating at present to propose the elimination of *Podoclavella,* because their close observations have been limited to only the Japanese species. *Pycnoclavella* seems valid for its two peculiarities, formation of the incubatory pouch in the distal part of the oviduct and three attachment processes issued directly from the anterior end of the embryonal or larval trunk instead of being supported by a special anterior protuberance as in *Clavelina.* Fusion of the test between zooids may attain distally to the middle of the thorax in some species of *Clavelina,* in which zooids are generally born on the common basal mass or stem, however no record of protrusion of the whole thorax above the common test has been made yet in *Synclavella*: this is the only reason why the present authors still admit *Synclavella* as a subgenus of *Clavelina.*

As already mentioned above, the following 7 clavelinids have been described from the Japanese waters.

1. *Dendroclavella elegans* Oka, 1927. Sagami Bay
   as *Clavelina fasciculata* Van Name, 1945
   Tokioka (1953): Ascidians of Sagami Bay.
2. *Clavelina coerulea* Oka, 1934. Kagosima and Mié Prefectures
3. *Podoclavella polycitorella* Tokioka, 1954. Tokara Islands
   Tokioka (1954): Publ. Seto Mar. Biol. Lab., vol. 3, no. 3, 250–251.
4. *Clavelina minuta* Tokioka, 1962. Sagami Bay
   Tokioka (1962): Publ. Seto Mar. Biol. Lab., vol. 10, no. 2, 272–274.
5. *Clavelina miniata* Watanabe and Tokioka, 1973. Near Simoda in Sagami Bay
   Watanabe and Tokioka (1973): Publ. Seto Mar. Biol. Lab., vol. 21, no. 2, 99–107.
6. *Clavelina cyclus* Tokioka and Nishikawa, 1975. Okinawa

Tokioka and Nishikawa (1975): Publ. Seto Mar. Biol. Lab., vol. 22, no. 5, 326–331.

7. *Clavelina obesa* n. sp.

In identifying the preserved specimens, probably the structures of the thorax may be the most reliable clues, especially the thoracic musculature and the ciliated groove are so in more or less contracted state. Of course, it is requested to learn how the musculature looks differently with the degree of contraction. Other important characters are mentioned in the key to Japanese species given next.

Key to the preserved specimens of the clavelinids so far described from the Japanese waters

Aa — Attached to the substratum quite independently by the ventral side of zooid, stomach surface smooth. ..... *Podoclavella polycitorella*

Ab — Attached to the common basal mass, stem or stolon by the posterior end of zooid. ..... B

Ba — Colony fasciate, zooids born on common stem. ..... *Clavelina elegans*

Bb — Zooids born separately on common basal mass or connected one another by stolons. C

Ca — Stigmatal rows 4, zooids connected by stolons. ..... *Clavelina minuta*

Cb — Stigmatal rows more than 10. ..... D

Da — Stomach near the posterior end of abdomen and folded very distinctly, stigmatal rows 10. ..... *Clavelina miniata*

Db — Stomach around the middle of abdomen, stigmatal rows more than 14. ..... E

Ea — A few longitudinal muscles crossing more than 20 transverse thoracic muscles, stigmatal rows 16–18, ciliated groove transverse. ..... *Clavelina coerulea*

Eb — Only oblique-longitudinal or -transverse thoracic muscles, ciliated groove longitudinal. ..... F

Fa — About 15 oblique-longitudinal thoracic muscles, 15–22 stigmatal rows. ..... *Clavelina cyclus*

Fb — About 10 nearly transverse thoracic muscles, 14–15 stigmatal rows. ..... *Clavelina obesa*

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EXPLANATION OF PLATE I

Above: *Clavelina coerulea* Oka on the southeastern coast of Mié Prefecture, in spring of 1975. By the courtesy of Mr. S. Shirai.

Below: *Clavelina cyclus* Tokioka and Nishikawa off Tinen, Okinawa Prefecture, in spring of 1975. By the courtesy of Mr. S. Shirai.